Studies of retinoids in the prevention and treatment of cancer

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Investigation of retinoids for anticancer activity in humans, either in the chemopreventive or treatment mode, has been little studied. We summarize here our ongoing investigations in four different areas: (1) secondary prevention of cervical dysplasia with topical application of all-*trans*-retinoic acid; (2) adjuvant treatment of resected high-risk stage I and II malignant melanoma with bacille Calmette Guérin (BCG) plus or minus oral vitamin A; (3) topical vitamin A acid therapy for cutaneous metastatic melanoma; and (4) oral isotretinoin as an anticancer agent. (J AM ACAD DERMATOL **6**:824-827, 1982.)

A considerable volume of information has been presented at this workshop which substantiates the therapeutic effect of synthetic retinoids on keratinizing disorders, acne, and basal cell epitheliomas. Extensive data in animals indicate that retinoids can suppress the phenotypic expression of epithelial malignancies, whether initiated by chemical or physical carcinogens.[1,2] Additionally, retinoids exert an antiproliferative effect against many normal and cultured cells[3] and in some instances promote differentiation,[4–6] a phenomenon best studied in murine[4] and human[5] melanoma and teratocarcinoma[6] cells. Investigations in Europe have also indicated that retinoids can inhibit the local growth of squamous cell malignancies of the skin.[7] We report here investigations which we have performed with retinoids in several different areas of clinical oncology. Four investigations will be presented and include:

1. Secondary prevention of cervical dysplasia with topical application of all-*trans*-retinoic acid (tretinoin) via a mechanical delivery system
2. Adjuvant treatment of resected high-risk stage I and stage II malignant melanoma with bacille Calmette Guérin (BCG) plus or minus oral vitamin A palmitate
3. Topical tretinoin therapy for cutaneous metastatic melanoma
4. Oral 13-*cis*-retinoic acid (isotretinoin) as an anticancer agent.

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Secondary prevention of cervical neoplasia with topical application of tretinoin

Vitamin A deficiency has long been known to be a cause of reversible epithelial dysplasia and metaplasia.[8] A considerable amount of epidemiologic data suggests that normal serum vitamin A levels protect against the development of epithelial malignancies.[9] The development of synthetic retinoids with minimal hepatic toxicity has led to extensive discussions concerning pharmacologic intervention in man in the chemopreventive mode.[10] What has not been adequately appreciated in these debates is the differences between primary, secondary, and tertiary prevention (Table I). Clearly, primary prevention may require only dietary supplementation with vitamin A, in most cases without active pharmacologic inter-

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vention. In contrast, secondary prevention will require the administration of pharmacologic doses of retinoids. It is also important to distinguish between those preneoplastic conditions which can be managed by local treatment versus those which would require systemic treatment. Examples of some preneoplastic states requiring local or systemic approaches are summarized in Table II. A carefully conducted investigation of retinoids as an antipromoter in the prevention of either oral, bladder, or cervical cancer could serve as a model for the use of retinoids against preneoplasia in humans. The regional administration of retinoids to the bladder or oral mucosa would be restricted to short-term administration due to the inability to conveniently deliver the compound under study continuously via local means. In contrast, local retinoid applications to the cervix via a mechanical delivery system should be possible (vide infra).

To administer a chemopreventive agent effectively to patients with cervical preneoplasia, several problems must be addressed: (1) choice of agent, (2) delivery of retinoid, (3) local and systemic toxicity of the compound, and (4) effectiveness. The choice of retinoid cannot be easily extrapolated from animal investigations since the results in preclinical studies are based on investigations of preneoplastic lesions of diverse histologic subtypes[11] and, furthermore, no animal model for cervical preneoplasia exists. We have selected tretinoin for several reasons: this retinoid has been used for many years for local control of acne, and a considerable amount of information is available regarding local skin and oral mucosa toxicity.[12] Additionally, well-defined liquid, gel, and cream vehicles are available.

The means of delivery to the cervix is critical since it is clear from animal studies that frequent, constant, and probably long-term application of the retinoid will be required to effect suppression of preneoplasia. Dr. Milos Chvapil, at the Univeristy of Arizona, developed a diaphragm lined with a collagen sponge which serves as a vehicle for the constant delivery of drugs to the cervix.[13] Using this delivery system, we will deliver tretinoin for a total of 4 days with daily replacement. In the initial studies, evaluation of toxicity will be important. We have therefore designed careful

Table I. Types of chemoprevention

Type	Example
Primary	Vitamin A deficiency
	Selenium deficiency
Secondary	Cervical dysplasia
	Squamous metaplasia of lung
Tertiary	"Surgically cured" tumor

Table II. Secondary prevention in preneoplasias

Local control
Cervical dysplasia
Oral leukoplakia
Bladder dysplasia
Lung dysplasia
Systemic control
Genetic syndromes
Preleukemia

criteria for toxicity, and escalation to the next dosage level will be done systematically. Our pilot investigations suggest that upper vaginal wall rather than cervical toxicity will be the dose-limiting side effect. Efficacy has not yet been determined. These studies will provide guidelines and have important implications for secondary prevention of other neoplasms.

Adjuvant treatment of stage I and stage II melanoma with oral vitamin A palmitate

The suppression or ablation of residual malignant disease after surgical or radiation therapy can be viewed as either prevention of recurrence or treatment of residual malignant cells. The treatment of recurrent melanoma is poor at best, and therefore considerable attention has been focused on adjuvant treatment, particularly with BCG and other immunomodulating agents. For a variety of reasons detailed elsewhere,[14] we initiated a stratified randomized study of BCG ± high-dose vitamin A in stage I and stage II malignant melanoma in April, 1978. Patients with histologically proved stage I and stage II melanoma were eligible. They were stratified by age, sex, and stage and randomized equally to receive either weekly Connaught BCG by scarification or BCG plus 100,000 units of vitamin A per day orally for a total of 18 months. Although the investigation has

not yet matured, an evaluation of the first forty-nine patients on study for at least 6 months has revealed that:

1. The relapse rate (6/21 in BCG group) (4/29 in BCG + vitamin A group) and pattern of recurrence (distant relapses in two of six patients on BCG, but 0 of 4 in BCG + vitamin A group) may indicate an advantage to patients receiving vitamin A. The difference in relapse-free survival in the two groups was not yet statistically significant ($p = 0.27$) but was encouraging since theoretically vitamin A, functioning as a hormone, may have been expected to induce melanoma growth.
2. Vitamin A was well tolerated and no evidence of liver damage was detected, as assessed by serial physical examinations, liver function studies, and liver scans.

Topical vitamin A acid for cutaneous malignant melanoma

Other investigations have demonstrated that application of tretinoin to squamous epithelial malignancies of the skin frequently resulted in regression.[7] We recently had the opportunity of treating two patients with cutaneous metastatic melanoma.[15] The patients were instructed to apply one drop of tretinoin (0.05% Retin-A solution) to each lesion once a day, and then the lesion was covered with occlusive tape. In one patient, complete clinical and histologic regression of twenty-one cutaneous lesions 0.5 to 1.0 cm in diameter was obtained. The patient remained free of disease for over 18 months from the start of therapy and for 12 months after the discontinuation of all therapy. He subsequently underwent relapse, with subcutaneous nodules in the previously diseased area. In the second patient, only two of twenty-two lesions responded clinically, and epidermal necrosis with acute inflammation with residual malignant cells was noted. The mechanisms producing these cellular responses to vitamin A acid are unknown, but these preliminary studies suggested that retinoids should be further examined for antitumor activity.

Antitumor activity of isotretinoin

Retinoids have classically been viewed as chemopreventive rather than as treatment modalities. However, retinoids clearly can inhibit malignant cells in culture[3] as well as inhibit human tumor stem cells[16] in a system which has been shown to be clinically predictive.[17] We therefore initiated a broad phase II anticancer trial of oral isotretinoin (3 mg/kg daily). The detailed results are presented elsewhere[18] and are summarized here. No activity was found against nonepithelial malignancies (0/21 cases), nor against nonsquamous epithelial cancers (one response in forty-five patients). One of thirteen patients with metastatic melanoma responded. In six of twenty-four patients with advanced squamous epithelial malignancies subcutaneous and skin disease sites responded. Remarkably, no bone marrow depression was seen, and skin toxicity was the limiting side effect.

These preliminary results give considerable impetus to further investigate the activity of isotretinoin and other retinoids against squamous epithelial malignancies and give merit to their utilization in the chemopreventive setting.

Future explorations of retinoids against advanced cancer

To utilize retinoids effectively as chemopreventive and/or therapeutic agents, several major avenues of investigations should be pursued:

1. What is the best way to deliver retinoids or other antipromoters? What is the safest solvent system? A large number of studies suggest that low doses of retinoids usually are antipromoters, but if given at high doses or with certain solvents, promotion may be dominant.*
2. What are the mechanisms by which the anticancer effect is achieved? Is it immunologic, membrane-mediated, via cellular retinoid acid binding protein?
3. What is the range of activity of retinoids in man? The recent availability of an in vitro assay system which predicts clinical efficacy with high accuracy for cytotoxic drugs may have a useful role in the screening of retinoids[16,17] and should decrease the amount of time and effort required to identify anticancer activity.
4. Can retinoids potentiate cytotoxic or hormonal drug action? Investigation of this neglected area is

*Slaga T: Personal communication.

needed, particularly since a few studies suggest potential synergisms.

Our exploration of retinoids as chemopreventive and treatment modalities is clearly in its infancy. These compounds represent one of the more well-defined areas of biologic modifiers, and an understanding of the mechanisms of action and clinical efficacy of retinoids should provide important clues to the noncytotoxic control of cancer.

A large number of colleagues are involved in these investigations, including Drs. D. Alberts, M. Chvapil, R. Dorr, W. Droegemueller, N. Levine, E. Surwit, and E. Gilmartin, R.N.

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